Test Definition: PLAFL

Platelet Surface Glycoprotein, Flow Cytometry, Blood

Overview

Useful For

Identification of markedly decreased CD41 (GPIIb) and CD61 (GPIIIa) expression levels, which are diagnostic for Glanzmann thrombasthenia

Identification of markedly decreased CD42a (GPIX) and CD42b (GPIb-alpha) expression levels, which are diagnostic for Bernard-Soulier syndrome

Identification of decreased GPVI expression, which suggests collagen receptor deficiency

Identification of decreased CD49b (GPIa), which suggests collagen receptor deficiency

Special Instructions

- Platelet Esoteric Testing Patient Information

Highlights

This test serves as a confirmatory test for platelet aggregation studies.

Markedly decreased platelet surface glycoprotein expression levels are diagnostic for various hereditary or acquired platelet disorders.

Method Name

Immunophenotyping

NY State Available

Yes

Specimen

Specimen Type

Whole Blood ACD

Ordering Guidance

This test identifies platelet surface glycoprotein or receptor deficiencies. If platelet structural abnormality testing is needed, consider PTEM / Platelet Transmission Electron Microscopic Study, Whole Blood.

Shipping Instructions

Specimen must be shipped ambient and arrive within 4 days of collection.

Ship specimen overnight in an Ambient Shipping Box-Critical Specimens Only (T668) following the instructions in the mailer.

Necessary Information

Platelet Esoteric Testing Patient Information **is required**. Testing may proceed without this information, however, the information aids in providing a more thorough interpretation. Ordering providers are strongly encouraged to fill out the form and send with the specimen.

Specimen Required

Supplies: Ambient Shipping Box-Critical Specimens Only (T668)
Collection Container/Tube: ACD solution A or B
Specimen Volume: 6 mL
Pediatric Volume: 1 mL
Collection Instructions: Send whole blood specimen in original tube. **Do not** aliquot.

Forms

1. Platelet Esoteric Testing Patient Information **is required**.
2. If not ordering electronically, complete, print, and send a Coagulation Test Request (T753) with the specimen.

Specimen Minimum Volume

Adult: 1 mL; Pediatric: 200 mcL

Reject Due To

Gross hemolysis	Reject
Fully clotted	Reject
Gross lipemia	OK

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Whole Blood ACD	Ambient	4 days	

Clinical & Interpretive

Clinical Information

Platelets have essential roles in primary hemostasis. Exposed collagen at a vascular damage site can activate platelets via collagen receptor GPVI and GPIa and bind shear-stretched multimeric VWF proteins, which subsequently interact with the platelet surface receptor, GPIb-V-IX. Upon full activation, platelets can aggregate by binding to fibrinogen through activated GPIIb-GPIIIa receptors. Deficiency of platelet surface glycoproteins can cause bleeding diathesis.

Platelet flow cytometric analysis is the preferred method to assess hereditary platelet disorders due to quantitative

surface glycoprotein (GP) deficiencies. GP expression levels can be measured by using fluorescent-conjugated GP-specific antibodies and their fluorescent intensities can be compared to normal ranges of various glycoproteins.

CD number	Glycoprotein name	Integrin name
CD41	GPIIb	Alpha 2b
CD42a	GPIX	NA
CD42b	GPIb-alpha	NA
CD49b	GPIa	Alpha 2
CD61	GPIIIa	Beta 3
NA	GPVI	NA

Reference Values

GPIIb CD41: > or =70.0% (Normal Range-Median)
GPIIIa CD61: > or =70.0% (Normal Range-Median)
GPIX CD42a: > or =70.0% (Normal Range-Median)
GPIb-alpha CD42b: > or =70.0% (Normal Range-Median)
GPIa CD49b: > or =60.0% (Normal Range-Median)

Interpretation

CD markers	% Reference range median	Comments
CD41 and CD61	50%-69% (Marginally)	Marginally decreased platelet surface receptors CD41 (GPIIb) and CD61 (GPIIIa) are of uncertain clinical significance. This finding could be a laboratory artifact due to a suboptimal sample condition, benign polymorphisms, or a heterozygous state of Glanzmann thrombasthenia. Recommend correlation with patient's clinical findings and results of platelet functional studies. Consider repeating platelet glycoprotein profile studies by flow cytometry to verify the present finding if clinically indicated.
	30%-50% (Moderately) <30% (Markedly)	Platelet surface expression of CD41 (GPIIb) and CD61 (GPIIIa) are moderately or markedly decreased. This finding is suggestive of a variant of Glanzmann thrombasthenia. Recommend correlation with patient's clinical findings and results of platelet functional studies. Consider repeating platelet glycoprotein profile studies by flow cytometry to verify the present finding if clinically indicated.
CD42a and CD42b	50%-69% (Marginally)	Marginally decreased platelet surface receptors CD42a (GPIX) and CD42b (GPIb-alpha) are of uncertain clinical significance. This finding could be a laboratory artifact due to a suboptimal sample condition, benign

		polymorphisms, or a heterozygous state of Bernard-Soulier syndrome. Recommend correlation with patient's clinical findings and results of platelet functional studies. Consider repeating platelet glycoprotein profile studies by flow cytometry to verify the present finding if clinically indicated.
	30%-50% (Moderately) <30% (Markedly)	Platelet surface expression of CD42a (GPIX) and CD42b (GPIb-alpha) are moderately or markedly decreased. This finding is suggestive of a variant of Bernard-Soulier syndrome. Recommend correlation with patient's clinical findings and results of platelet functional studies. Consider repeating platelet glycoprotein profile studies by flow cytometry to verify the present finding if clinically indicated.
CD49b	30%-59% (Marginally)	Marginally decreased platelet surface receptor CD49b (GPIa) is of uncertain clinical significance. This finding could be a laboratory artifact due to a suboptimal sample condition, a benign polymorphism, or a variant of platelet collagen receptor glycoprotein Ia/IIa deficiency. Recommend correlation with patient's clinical findings and results of platelet functional studies. Consider repeating platelet glycoprotein profile studies by flow cytometry to verify the present finding if clinically indicated.
	10%-30% (Moderately) <10% (Markedly)	Platelet surface expression of CD49b (GPIa) is moderately or markedly decreased. This finding is suggestive for a variant of a variant of platelet collagen receptor glycoprotein Ia/IIa deficiency. Recommend correlation with patient's clinical findings and results of platelet functional studies. Consider repeating platelet glycoprotein profile studies by flow cytometry to verify the present finding if clinically indicated.
GPVI	50%-69% (Marginally)	Marginally decreased platelet surface receptor glycoprotein VI (GPVI) is of uncertain clinical significance. This finding could be a laboratory artifact due to a suboptimal sample condition, a benign polymorphism or a variant of platelet collagen receptor GPVI deficiency. Recommend correlation with patient's clinical findings and results of platelet functional studies. Consider repeating platelet glycoprotein profile studies by flow cytometry to verify the present finding if clinically indicated.
	30%-50% (Moderately)	Platelet surface expression of glycoprotein VI (GPVI) is moderately or markedly decreased. This finding is

	<30% (Markedly)	suggestive of a variant of a variant of platelet collagen receptor GPVI deficiency. Recommend correlation with patient's clinical findings and results of platelet functional studies. Consider repeating platelet glycoprotein profile studies by flow cytometry to verify the present finding if clinically indicated.

Cautions

Suboptimal sample conditions due to improper blood draw, transportation, or storage may cause fluctuation of platelet surface receptors and consequently influence the results of platelet surface receptor measurement by flow cytometry.

Supportive Data

The platelet glycoprotein flow cytometry method was established in the Mayo Clinic Special Coagulation Laboratory in 2009. Between 2009 to 2014, a total of 155 clinical patients at Mayo Clinic were tested. The flow cytometry results were compared with the final impressions of platelet light transmission aggregation testing. There were 7 samples that had flow cytometric features of Glanzmann thrombasthenia, 2 samples that had flow cytometric features of Bernard-Soulier syndrome, and 3 samples that had flow cytometric features of May-Hegglin anomaly. All flow cytometric results were concordant with platelet light transmission aggregation results and other clinical findings.

Clinical Reference

1. Miller, JL. Glycoprotein analysis for the diagnostic evaluation of platelet disorders. Semin Thromb Hemost. 2009;35(2):224-232
2. Kannan M, Ahmad F, Yadav BK, et al. Carrier detection in Glanzmann thrombasthenia: comparison of flow cytometry and Western blot with respect to DNA mutation. Am J Clin Pathol. 2008;130(1):93-98
3. Savoia A, Pastore A, De Rocco D, et al. Clinical and genetic aspects of Bernard-Soulier syndrome: searching for genotype/phenotype correlations. Haematologica. 2011;96(3):417-423
4. Nurden AT, Freson K, Selifsohn U. Inherited platelet disorders. Haemophilia. 2012;18(4):154-160
5. Spurgeon BEJ, Naseem KM. Platelet flow cytometry: Instrument setup, controls, and panel performance. Cytometry B Clin Cytom. 2020;98(1):19-27
6. Frelinger AL 3rd, Rivera J, Connor DE, et al. Consensus recommendations on flow cytometry for the assessment of inherited and acquired disorders of platelet number and function: Communication from the ISTH SSC Subcommittee on Platelet Physiology. J Thromb Haemost. 2021;19(12):3193-3202

Performance

Method Description

Flow cytometric immunophenotyping of peripheral blood platelets is performed using the following antibodies:

Panel: CD41 (IIb), CD42a (IX), CD42b (Ib-alpha), CD49b (GPIa), CD61 (GPIIIa), and GPVI.
For sample quality purposes, CD62P is evaluated.

Using whole blood collected in ACD (A or B), platelet surface GPIa, Ib-alpha, IIb, IIIa, VI and IX expression levels are

measured by flow cytometry method. Platelets in whole blood are stained with various fluorochrome-labeled primary antibodies and fixed. Then the platelet surface fluorescent intensities of various bound antibodies are measured by flow cytometers. Platelets are first gated by forward and side scatter. Mean fluorescent intensities are recorded and converted to percentage of a median fluorescent intensity of a normal donor study of 20 healthy donors. If the percentage of expression of a glycoprotein (GP) is lower than the corresponding normal range, a deficiency of a GP is detected.(Unpublished Mayo method)

PDF Report
No

Day(s) Performed
Monday through Saturday

Report Available
1 to 2 days

Specimen Retention Time
14 days

Performing Laboratory Location
Mayo Clinic Laboratories - Rochester Main Campus

Fees & Codes

Fees
- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

Test Classification
This test was developed and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. It has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information
88184-Flow cytometry; first cell surface, cytoplasmic or nuclear marker
88185-Flow cytometry; additional cell surface, cytoplasmic or nuclear marker (each) X5
88187-Flow cytometry interpretation, 2 to 8 markers

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
PLAFL	Platelet Glycoprotein Flow, B	93320-0

Result ID	Test Result Name	Result LOINC® Value

CK111	GPIIb CD41	93319-2
CK112	GPIIIa CD61	93318-4
CK113	GPIX CD42a	93317-6
CK114	GPIb-alpha CD42b	93316-8
CK115	GPIa CD49b	93315-0
CK116	GPVI	93314-3
CK117	Final Diagnosis	93313-5